

## PCR PRODUCT CONCENTRATION BY EVAPORATION

**Purpose:** There are times when it is necessary to adjust the relative concentrations of PCR products on a plate-wise basis prior to spotting. This is done by evaporation, rather than purification and re-suspension, since nylon membranes can accommodate low levels of reagent contamination. We believe this is quicker, less expensive, and in some cases, can provide a more uniform product than a protocol such as ethanol precipitation or column purification.

### Materials Needed:

- ☐ DEPC H<sub>2</sub>O
- ☐ Plate sealers (Edge Biosystems, cat# 48461)
- ☐ cyclofoil roller (Apogent Discoveries, cat# 1044-39-2)
- ☐ MJ Research Tetrad PCR machine (MJ Research, cat# PTC-225)

### Procedure:

1. Set PCR machine (MJ PTC-225) to run continuously at 72 degrees. Disable the heated lid.
2. Remove the plate sealers and insert the plates into the PCR machine, leaving the lids up.
3. Evaporate PCR product to approximately 100 ul.
4. After 15 minutes, flip the plates around to account for machine variability.
5. When evaporation is complete (approximately 30 minutes), remove the plates and let them cool to room temperature.
6. In cases of uneven evaporation, add 10 to 20 ul DEPC water to even the wells up.
7. Cover with plate sealers and roll with cyclofoil roller.
8. Store at -20 degrees. Plates are ready for preparation for arraying.

### Comments:

There is no general need to purify PCR products when spotting onto nylon membranes. The need to evaporate should be determined empirically relative to the average amount of DNA per well. For PCR runs of high efficiency there may be no need to concentrate. For lower efficiency runs, it may be necessary to concentrate the products.

### Contacts:

Kayris Wall	410-558 8300 X 7165	<a href="mailto:grayKa@grc.nia.nih.gov">grayKa@grc.nia.nih.gov</a>
William H. Wood III	410-558-8327	<a href="mailto:woodw@grc.nia.nih.gov">woodw@grc.nia.nih.gov</a>

For frequently asked questions go to the following address:  
<http://www.grc.nia.nih.gov/branches/rb/dna/protocolFAQs.htm>

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